

ORIGINAL ARTICLES

THE STUDY OF PHENOLIC COMPOUNDS IN CANNA LILY FLOWERS USING HPLC

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ABSTRACT

Aim: The purpose of our work was the research of phenolic compounds in canna lily (*Canna x hybrida* Hort.) flowers for a further detailed study of the phytochemical composition of raw material from the canna lily plant.

Materials and Methods: Canna lily flowers of red color collected in Ukraine in the summer seasons of the period 2014-2016 were chosen as the object of the study. Ethanol extracts obtained from dried flowers were used for the research. The extracts were analyzed using the high-performance liquid chromatography (HPLC) technique on a liquid chromatograph equipped with a diode-array detector Shimadzu HPLC-system, ser. 20. HPLC was used to detect phenolic compounds in canna lily flowers.

Results and Conclusion: Rutin, neochlorogenic, chlorogenic, caffeic and rosmarinic acids were identified in canna lily flowers.

The obtained experimental data can be used in the development of the standardization parameters of canna lily flowers.

Keywords: canna lily, rutin, neochlorogenic acid, chlorogenic acid, caffeic acid, rosmarinic acid, high performance liquid chromatography

INTRODUCTION

Carrying out studies of plants used in traditional medicine, which have vast raw material base, is important in order to expand the range of medicinal plant raw material at the modern pharmaceutical market.

Canna lily is one of these plants.

Canna lily is not an officinal plant in Ukraine. Nevertheless, canna lily is a prospective plant for a complex pharmacognostic study and further creation of new phytochemical remedies on its basis due to the diverse composition of biologically active compounds in its content.

From the literature it is known that other members of the *Cannaceae* family (including *Canna indica* L.) have been widely used in folk medicine as anti-inflammatory, immunomodulatory, antioxidant, antidiabetic, anticancer agents for a long time (1,2). Phenolic biologically active substances correspond with these types of activity (3-5). Hydroxycinnamic acids and flavonoids, including a flavonoid glycoside rutin, also show these types of pharmacological action (3,6-9). Therefore a study of phenolic compounds in canna lily flowers is a basis for their further implementation in medical practice.

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AIM

The purpose of this work was a study of phenolic compounds in canna lily flowers for a more detailed research on phytochemical composition of canna lily plant raw material.

MATERIALS AND METHODS

Ethanol extract preparation:

The determination was carried out by the high-performance liquid chromatography (HPLC) technique applying the following protocol: 0.50g (exact weight) of the minced plant raw material was put into a 100-ml conical flask with reflux condenser followed by the addition of 25ml of 50% ethanol. The flask was left in a boiling water bath for 45min. After this the extract was cooled to room temperature and was filtered through the “red stripe” filter into a volumetric flask with a volume of 25ml. The extract volume was adjusted till the mark with 50% ethanol. The chromatographic determination of phenolic compounds was carried out on a liquid chromatograph equipped by diode-array detector Shimadzu HPLC-system, ser.20 in the following conditions: Phenomenex Luna C18(2) column. Column size was 250mm x 4.6mm with particle size of 5µm and column temperature – 350°C. The detection wavelength equaled 330 nm, mobile phase flow rate was 1ml/min; the introduced

sample volume was 5µl. The mobile phase was the following: Chromatography time (min) Eluent A, % Eluent B, % 0–5 95 5 5–35 95 → 75 5 → 25 35–40 75 25 40–60 75 → 50 25 → 50 60–65 50 → 20 50 → 80 65–70 20 80 70–85 95 5; Eluent A: 0.1% solution of trifluoroacetic acid in water; Eluent B: 0.1% solution of trifluoroacetic acid in acetonitrile. Identification of components was carried out by retention time and UV spectra comparison in accordance with standards.

RESULTS AND DISCUSSION

According to the analysis results, rosmarinic, gallic and caffeic acids were identified with the help of HPLC. Their content was determined in the samples of canna lily plant raw material. The result of the study is presented in Figure 1 and Table 2.

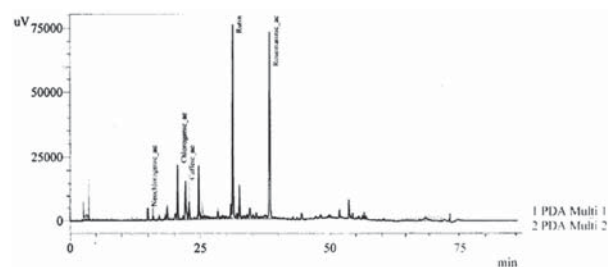


Fig. 1. Chromatogram of the phenolic compounds of canna lily flowers without hydrolysis

Table 1. Mobile phase

Time, minutes	Mobile phase A, % (turnover/turnover)	Mobile phase B, % (turnover/turnover)	Mode
0→1	92	8	isocratic
1→15	92→70	8→30	gradient
15→25	70→0	30→100	gradient
25→33	0	100	isocratic
33→33.5	0→92	100→8	gradient

Table 2. Phenolic compounds' content in canna lily plant raw material

Nº	Retention time, min	Phenolic acids	Formula	Molecular mass g/mol	Content in a studied sample, %
1	14.950	Neochlorogenic acid	C16H18O9	354.31	Trace amount
2	20.647	Chlorogenic acid	C16H18O9	354.31	0.0367
3	22.164	Caffeic acid	C9H8O4	180.16	0.0204
5	38.270	Rosemarinic acid	C18H16O8	360.31	0.1409
4	31.200	Rutin	C27H30O16	610.52	0.2415

Rutin, neochlorogenic, chlorogenic, caffeic and rosmarinic acids were identified in canna flowers. Rutin was a dominant compound among detected phenolic compounds. Its content equaled $0.2415 \pm 0.0100\%$. Rosemarinic acid was a dominant component among the determined phenolic acids. Its content equaled $0.1409 \pm 0.0200\%$. Chlorogenic and caffeic acids were found in smaller amounts and their content was $0.0367 \pm 0.0100\%$ and $0.0204 \pm 0.0100\%$, respectively. Only trace amounts of neochlorogenic acid were found.

CONCLUSION

Currently, there is an increasing demand for drugs obtained from medicinal plant raw material in the pharmaceutical market of Ukraine as well as abroad.

The conducted study allows the development standardization methods for canna lily plant raw material according to the following data:

1. Five phenolic compounds, such as rutin, neochlorogenic, chlorogenic, caffeic and rosmarinic acids were determined in canna lily flowers by the HPLC technique.
2. Rutin dominated by content the other phenolic compounds identified. Its amount equaled $0.2415 \pm 0.0100\%$.
3. Rosemarinic acid was a dominant component by content among the phenolic acids determined. Its content equaled $0.1409 \pm 0.0200\%$. Chlorogenic and caffeic acids were found in smaller amounts and their content was $0.0367 \pm 0.0100\%$ and $0.0204 \pm 0.0100\%$, respectively. Only trace amounts of neochlorogenic acid were found.
4. The obtained experimental data can be used in the development of standardization parameters for canna lily flowers as well as for the subsequent development of substances from the analyzed plant raw material.

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